EZBiolab

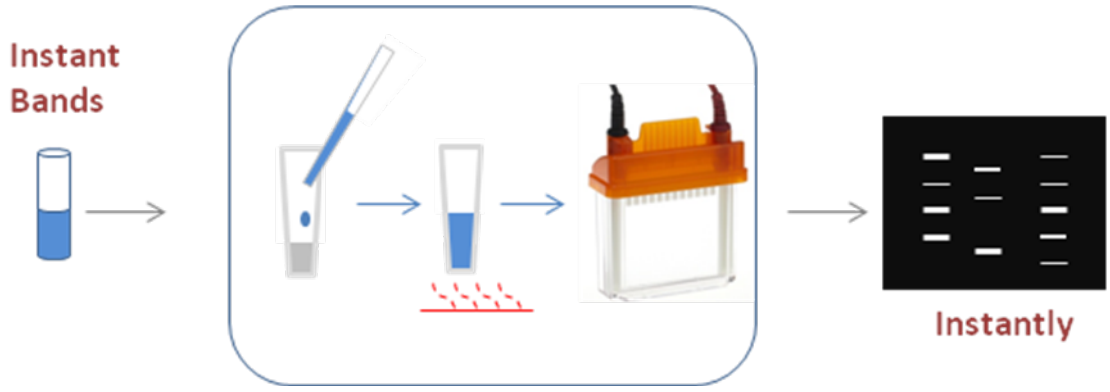
View Your SDS-PAGE Results Instantly, No More Gel Stain and De-Stain

User's Manual

Instant-Band sample treatment buffer (sample loading buffer) pre-stains a protein sample for SDS-PAGE. Protein samples are stained fluorescently during sample treatment prior to electrophoresis. You can observe results directly after electrophoresis on an UV or LED blue transilluminator. This product is

- **Simple:** Completely eliminate the stain-destain procedure without introducing any extra steps
- **Fast:** Protein bands in the gel can be visualized and pictured right after electrophoresis
- **Sensitive:** The sensitivity of detection of proteins is higher than the conventional Coomassie blue
- **Convenient:** The Gel doesn't need to be removed from the gel cassettes when using a LED transilluminator
- **Flexible:** The gel pre-stained with Instant-Band can still be processed by Coomassie Blue staining
- **Clean:** No contamination of organic solvents to our environment and no mess of Coomassie Blue to your lab

Experiment Scheme of Instant-Band



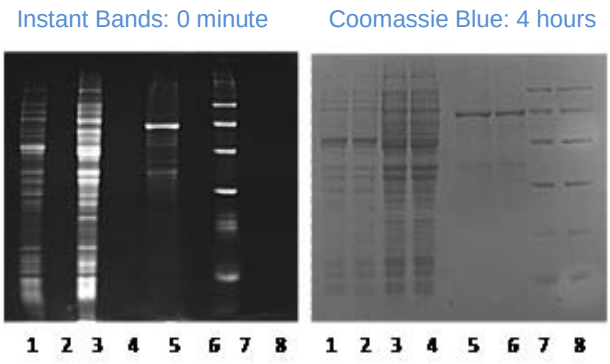
Product

Product	Catalog #	Package	Amount*
Instant-Band Protein Sample Treatment Buffer for SDS-PAGE	PFS001	2 x Instant-Bands (1 ml/tube)	> 800 lanes

* 5 ul loading volume each lane

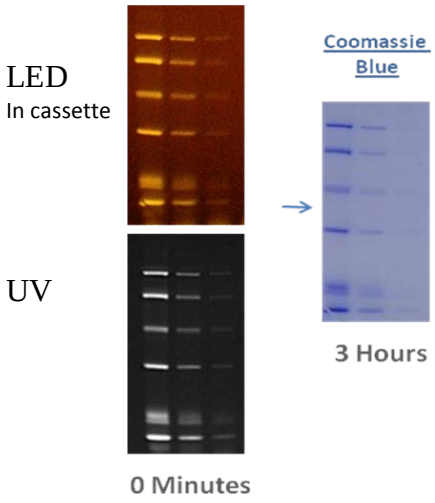
Directions

1. Mix Instant-Band Sample Treatment Buffer with a protein sample in 1:1 ratio.
2. Heat the sample at 90-100 ºC for 10 minutes. Make sure your heating block is >90ºC to let the sample be sufficiently heated.
3. The sample is ready for gel electrophoresis.
4. After electrophoresis, place the gel on an UV or LED blue transilluminator to view protein bands and take pictures. Because light emitted by LED is transmittable through glass or plastics, the gel does not need to be removed from the gel cassette.



1, 3, 5 and 7 were treated by Instant-Band loading buffer

2, 4, 6 and 8 were treated by the regular loading buffer



Storage and Stability

- Immediately store the package at -20 ºC upon receiving.
- Instant-Band Sample treatment Buffer is stable for 12 month at -20 ºC, 8 weeks at 4 degree and 1 week at room temperature.
- Like regular sample treatment buffer, oxidation of reducing reagent such as DTT is the major problem for long term storage of Instant-Band at room temperature. Some protein bands could become less sharp and less bright if Instant-Band is stored at RT for more than a week. Add freshly made DTT will restore band sharpness and brightness. The final concentration of DTT in Instant-Band should not be over 20 mM
- Always avoid light.

Additional Features

1. Gels stained by Instant-Band can still be stained by Coomassie blue if it is wanted. Perform Coomassie blue stain experiments according to the standard protocol.
2. Instant-Band is compatible to Western Blot. Western Blot experiments can be performed using the standard protocol. Proteins transferred onto the blot membrane can be directly visualized before and after antibody binding, which helps to identify the target proteins. Alternatively, Instant-Bands stained samples can be run as a control lane to verify transfer efficiency of the target proteins.

Conditions and Factors

M.W. Marker Instant-Band is specially formulated to enhance its stain efficiency. Most of ready-use protein molecular weight markers are not compatible with Instant-Band. EZBiolab provides pre-stained fluorescent protein M.W. markers. Alternatively, markers can be generated by staining untreated proteins with Instant-Bands.

Effect of pH Low pH reduces stain efficiency. If pH of a sample solution is below 5, we suggest you to adjust pH to above 7 for the best results. High pH (>7) does not affect Instant-Band.

Other Factors Proteins are effectively stained by Instant-Bands under normal conditions. Table below summarizes the effect of common reagents found in protein solution (Control is 5+)

Detergents		0.1 %	0.5%	1%	
	Triton - 100	5 +	5 +	5 +	
	Chaps	5 +	5 +	5 +	
	Glycoside	5 +	5 +	5 +	
	Tween	5 +	5 +	5 +	
Buffers		25 mM	50 mM	100 mM	
	Pi Buffer, pH7.4	5 +	5 +	5 +	
	Tris Buffer, pH 7.4	5 +	5 +	3 +	
	PIPES Buffer, pH 6.5	5 +	4 +	2 +	
	HEPES Buffer, pH 7.5	5 +	5 +	4 +	
Others	DTT	10 mM	20 mM	30 mM	50 mM
		5 +	5 +	5 +	4 +
	DMSO	1%	3%	5%	
		5 +	5 +	5 +	
	Urea	2M	4 M	6 M	8 M
		5 +	5 +	4 +	3 +
	Glycerol	5%	10%	20%	
		5 +	4 +	3 +	

For protein under extreme conditions such as high concentration of urea or low pH (<5), please use Enhanced Instant-Band.

Application

The product is best used for SDS-PAGE electrophoresis to check protein expression, determine the purity of a protein sample, track target protein during purification and western blot as well as to exam immune-precipitation results

The product is not recommended for experiments to cut protein bands from a gel for sequencing, mass spectrometry analysis or antibody production.

Frequent Asked Questions and Trouble Shooting

1. Why can't ready-to-use protein M.W. markers I purchased from other vendors be stained by Instant-Band effectively?

Instant-Band is specially formulated to enhance its staining efficiency. Most of the ready-use protein molecular weight markers are not compatible with Instant-Bands. EZBiolab provides pre-stained fluorescent protein M.W. marker. Alternatively, markers can be generated by staining untreated proteins with Instant-Bands.

2. How about the stability of Instant-Band?

Instant-Band dye itself is stable. However, same as the regular sample treatment buffer, oxidation of reducing reagent such as DTT could reduce sharpness and brightness of some protein bands. The recommended storage time of instant-Band loading buffer at room temperature is one week. If more than one week, adding freshly made DTT to Instant-Band will restore the sharpness and brightness. The final concentration of DTT in Instant-Band should not be over 20 mM DTT. Other reducing reagents such as TCEP (2 mM) will also work well.

3. Do post-translational modifications affect Instant-Bands?

No

4. Some protein bands get less sharp over time, why?

Oxidation of protein reduces band sharpness. Add freshly made DTT to Instant-Bands. DTT concentration should not be over 20 mM.

5. Can I do Western Blot experiments using Instant-Bands stained samples?

Yes. Instant-Bands do not affect Western Blot results. Alternatively, you can use Instant-bands stained sample as a control lane to check expression level of the target protein and verify the transfer efficiency.

6. Why do I see precipitation in Instant-Bands that has been stored at 4 degree?

The precipitation is SDS. It will disappear when temperature is raised to at room temperature.

Related Products

- Fluorescent Protein M.W. Marker
- EZ-Viewer LED Transilluminator
- EZ-PAGE, the next generation of precast gel